ORIGINAL ARTICLE

Evaluating the drivers of Indo-Pacific biodiversity: speciation and dispersal of sea snakes (Elapidae: Hydrophiinae)

Kanishka D. B. Ukuwela[1,2], Michael S. Y. Lee[1,3], Arne R. Rasmussen[4], Anslem de Silva[5], Mumpuni[6], Bryan G. Fry[7], Parviz Ghezellou[8], Mohsen Rezaie-Atagholipour[9] and Kate L. Sanders[1]*

[1]School of Biological Sciences, University of Adelaide, Darling Building, Adelaide, SA 5005, Australia, [2]Department of Biological Sciences, Faculty of Applied Sciences, Rajarata University of Sri Lanka, Mihintale 50300, Sri Lanka, [3]Earth Sciences Section, South Australian Museum, North Terrace, Adelaide, SA 5000, Australia, [4]Schools of Architecture, Design and Conservation, The Royal Danish Academy of Fine Arts, Esplanaden 34, Copenhagen K. DK-1263, Denmark, [5]Amphibian & Reptile Research Organization of Sri Lanka, 15/1, Dolosbage Rd., Gampola, Sri Lanka, [6]Museum of Zoology Bogor, Puslit Biology-LIPI, Cibinong, Indonesia, [7]Venom Evolution Laboratory, School of Biological Sciences, University of Queensland, Brisbane, QLD 4072, Australia, [8]Department of Phytochemistry, Medicinal Plants and Drugs Research Institute, Shahid Beheshti University, G.C. Evin, P.O. Box 19835-389, Tehran, Iran, [9]Environmental Management Office, Qeshm Free Area Organization, Qeshm Island, Hormozgan Province, Iran

ABSTRACT

Aim There are several competing hypotheses to explain the high species richness of the Indo-Australian Archipelago (IAA) marine biodiversity hotspot centred within Southeast (SE) Asia. We use phylogenetic methods to provide a novel perspective on this problem using viviparous sea snakes, a group with high species richness in the IAA that is highly distinct from other taxa previously studied, both phylogenetically (Reptilia, Amniota) and biologically (e.g. viviparity and direct development).

Location Indian Ocean and the West Pacific.

Methods We used likelihood and Bayesian methods to reconstruct a multi-locus time-calibrated phylogeny for *c.* 70% of viviparous sea snake species, many sampled from multiple localities in Australasia, Southeast Asia and the Indian Ocean. We then compared rates and temporal concordance of inferred vicariance and dispersal events between marine basins using several approaches including new Bayesian analyses that allow for clade-specific and event-specific dispersal rates.

Results Phylogenetic analyses and novel Bayesian biogeographical reconstructions indicate that viviparous sea snakes underwent rapid speciation after colonizing SE Asia *c.* 3 million years ago. Most of the SE Asian sea snake diversity is the result of *in situ* speciation, most consistent with the 'centre of origin' and 'centre of refuge' models for biodiversity hotspots. There is also speciation at the periphery, or entirely outside SE Asia; however, contrary to predictions of the 'accumulation' and 'overlap' models, these new outlying taxa do not preferentially disperse back into SE Asia. Instead, lineages are equally likely to disperse either into or away from SE Asia.

Main conclusion The high diversity of sea snakes in SE Asia (and hence the IAA) is mostly explained by *in situ* speciation rather than accumulation or overlap. Most speciation events are contemporaneous with sea level changes that generated and dissolved barriers between marine basins during the last 2.5 million years.

Keywords
biodiversity hotspot, centre of origin, Coral Triangle, evolutionary radiation, Indo-Australian Archipelago, Pleistocene

*Correspondence: Kate L. Sanders, School of Biological Sciences, University of Adelaide, Darling Building, Adelaide, SA 5005, Australia
E-mail: kate.sanders@adelaide.edu.au

INTRODUCTION

The Indo-Australian Archipelago (IAA), situated between the Indian and Pacific Oceans (see Fig. S1 in Appendix S1, Supporting Information), supports an exceptionally rich concentration of marine biodiversity (Hughes *et al.*, 2002), with more fish and coral species reported than for any other region (Hoeksema, 2007; Allen, 2008). A pattern of declining diversity with latitudinal and longitudinal distance from the central IAA in many taxa (Veron, 1995; Briggs, 1999; Mora

et al., 2003) suggests that a common process underlies this biodiversity hotspot. Theories proposed to explain the exceptional IAA marine diversity typically view the region as either: (1) a centre of origin/speciation, where new species form rapidly and subsequently disperse to peripheral areas (Ekman, 1953); (2) a centre of accumulation of diversity, with speciation in isolated locations at the periphery of the IAA and subsequent movement of newly-formed taxa into the region (Ladd, 1960); or alternatively (3) a region of overlap for marine biodiversity that originated in the Pacific and Indian Oceans, i.e. completely outside the IAA (Woodland, 1983). Finally (4), the centre of refuge model suggests that the temporally stable habitat-rich IAA has enabled prolonged speciation and survival for marine species (Jackson *et al.*, 1993; Bellwood & Hughes, 2001; Pellissier *et al.*, 2014), with the diversity gradient due to elevated extinction outside the IAA.

Studies on various marine taxa from the region have provided support for the 'centre of origin' (Veron, 1995; Carpenter & Springer, 2005; Barber *et al.*, 2006; Tornabene *et al.*, 2015), 'centre of accumulation' (Drew & Barber, 2009; Eble *et al.*, 2011; Hodge *et al.*, 2012), 'region of overlap' (Santini & Winterbottom, 2002; Hubert *et al.*, 2012; Gaither & Rocha, 2013), and the 'centre of refuge' (Pellissier *et al.*, 2014) models. Taken together, these studies suggest that multiple processes could contribute towards higher IAA marine biodiversity in various taxa (Randall, 1998; Bernardi *et al.*, 2004; Barber & Bellwood, 2005; Mironov, 2006), and have led to a 'biodiversity feedback model' under which the IAA and other tropical marine biodiversity hotspots act as centres of speciation, accumulation and/or overlap (Bowen *et al.*, 2013).

Determining the relative importance of these potential processes generating IAA marine biodiversity requires study groups that span the Indo-Pacific, are highly species-rich, are relatively young (thus preserving recent biogeographical events) and can be well-sampled for phylogenetic analysis. The viviparous sea snakes (Elapidae: Hydrophiinae) offer high species diversity, with 62 described species that share a terrestrial Australian ancestor only *c.* 10.6–6.5 million years ago (Ma) (Sanders & Lee, 2008; Sanders *et al.*, 2008; Lukoschek *et al.*, 2012). They occupy shallow-marine habitats throughout the tropical and subtropical Indian and Pacific Oceans, but like many other marine groups in the Indo-Pacific, reach peak species diversity in the IAA hotspot (Elfes *et al.*, 2013). Moreover, at least 75% of sea snake species are part of a single, explosively speciating 'core *Hydrophis* clade', less than *c.* 3 million-years old (Sanders *et al.*, 2010, 2013a) and widespread throughout the IAA.

The majority of sea snake diversification, including the rapid core *Hydrophis* radiation, occurred during major climatic and geological events (Voris, 2000; Woodruffe, 2003; Sanders *et al.*, 2013a) that drove vicariant population and species divergence in many of the region's marine groups (reviewed in Carpenter *et al.*, 2011). Viviparous sea snakes might be particularly influenced by 'soft' biogeographical barriers (such as incomplete and thus permeable land bridges) because they undergo direct development (i.e. give birth to live young) and thus lack the dispersing planktonic larval stage that is expected to promote population connectivity in most other marine groups (many fish and invertebrates) (Hoskin, 1997). Several sea snake species accordingly show strong intraspecific genetic structure corresponding to deep-water and historical land barriers (Lukoschek *et al.*, 2007; Sanders *et al.*, 2013b; Ukuwela *et al.*, 2014). However, biogeographical patterns and the diversification dynamics of the entire sea snake radiation have not previously been quantitatively investigated.

In this study we aimed to resolve the biogeographical history of viviparous sea snakes using a multi-locus time-calibrated phylogeny for *c.* 70% of described species, many sampled from multiple localities. We then compared rates and temporal concordance of inferred vicariance and dispersal events between marine basins in Australasia, SE Asia and the Indian Ocean. Specifically, our objective was to test whether viviparous sea snake diversity in the IAA is best explained by *in situ* speciation, peripheral speciation and accumulation, or external speciation and subsequent overlap. We use several approaches including new Bayesian analyses that allow for clade-specific and event-specific dispersal rates.

Although numerous studies have investigated the biogeography of Indo-Pacific marine taxa, most of these have involved a single (Williams & Benzie, 1998; Gaither *et al.*, 2011) or a few species (Halas & Winterbottom, 2009; Gaither *et al.*, 2010), and many have been restricted to sub-regions/single marine basins (Barber *et al.*, 2000; Lourie & Vincent, 2004; Lukoschek *et al.*, 2007). The few broad scale biogeographical studies of species-rich, widely distributed groups have focused primarily on reef fish (Barber & Bellwood, 2005; Gaither & Rocha, 2013). Our study of sea snakes thus provides a novel insight towards understanding the biogeographical processes that have shaped this important marine region.

METHODS

Sampling

We sampled a total of 320 individuals from 42 species of viviparous sea snakes from Australia, Indonesia, Myanmar, Malaysia, Vietnam, Thailand, Bangladesh, Sri Lanka, India and Iran (see Fig. S1 in Appendix S1). The specimens were (provisionally) identified following descriptions and diagnoses of Smith (1926) and Rasmussen (2001); taxonomy and nomenclature follows Sanders *et al.* (2013a). Liver/muscle tissue samples preserved in 90% ethanol/isopropanol were obtained from specimens collected primarily as fisheries by-catch (233 individuals, 36 species) and from specimens accessioned in museums (57 individuals, 22 species). Additional mitochondrial and nuclear sequences were also obtained from Genbank (30 individuals, 16 species). Specimen collection localities and museum voucher numbers are provided in Appendix S2 (Supporting information).

We amplified and sequenced a total of 5792 base pairs (bp) from three mitochondrial markers (cytochrome *b* [*cyt b*]: 1095 bp, NADH dehydrogenase subunit 4 [*ND4*] and adjacent *tRNA* region: 838 bp, 16S small subunit of ribosomal RNA [*16SrRNA*]: 531 bp), two nuclear coding genes (Oocyte maturation factor [*c-mos*]: 918 bp, recombination activation gene [*RAG-1*]: 1066 bp) and three nuclear anonymous markers (*G1888*: 428 bp, *G1894*: 422 bp, *G1914*: 494 bp) to reconstruct sea snake phylogeny. Details of DNA extraction, PCR amplification and sequencing are available in Appendix S1. The sequences generated in this study are deposited in the Genbank sequence database (see Appendix S2 in Supporting Information).

Phylogeny and divergence time estimates

Time-calibrated sea snake phylogenies were inferred using maximum likelihood (ML) and Bayesian analyses of the concatenated mitochondrial and nuclear alignment (See Appendix S1 in Supporting Information for details). The Australasian terrestrial elapid *Hemiaspis damielli* was used as an outgroup because there is strong molecular and morphological evidence that *Hemiaspis* is a close relative of the viviparous sea snakes (= Hydrophiini) (Rasmussen, 2002; Lukoschek & Keogh, 2006; Sanders *et al.*, 2008). Maximum Likelihood analyses (undated, no clock) were implemented in RAxML 7.2.8 (Stamatakis, 2006). For the (dated) Bayesian analyses, Bayes Factors (ΔBF; *sensu* Kass & Raftery, 1995) strongly supported the strict clock over the uncorrelated gamma relaxed clock (ΔBF = 1938); this was consistent with undated (clock-free) trees being approximately ultrametric. The prior on overall rate was set to encompass a broad range, with a lower bound of zero and an upper 95% bound of 20% per lineage per million years, *c.* 20× the 'typical' rate of mtDNA (normal distribution with mean 0.01 substitutions per million years and a standard deviation of 0.1, truncated at 0). Bayesian analyses with estimation of the divergence times were performed in MRBAYES 3.2 (Ronquist & Huelsenbeck, 2003) (see Appendix S3 in Supporting Information for Nexus alignment with MRBAYES command block). As there are no known Hydrophiini fossils that could be used to calibrate the tree, secondary calibrations (uniform distributions 6.5–10.6 Ma and 4.5–7.9 Ma) were applied, respectively, to the root divergence and the *Aipysurus-Hydrophis* divergence. These bounds correspond to the 95% HPD distributions estimated for these two divergences in wider squamate analyses using long nuclear sequences and several reliable squamate fossil calibrations (Sanders *et al.*, 2008; Scanlon & Lee, 2011; Lukoschek *et al.*, 2012). Convergence of the independent runs in topology was assessed by examining similar clade (split) frequencies across runs (standard deviation < 0.05); convergence in numerical parameters was assessed although essentially identical distributions with high effective sample sizes (> 200) as shown by TRACER 1.5 (Rambaut & Drummond, 2007).

In addition to the phylogenetic analyses, we estimated the genetic distance between sister lineages in different ocean basins to examine the amount of genetic divergence between these distinct lineages: corrected Hasegawa-Kishino-Yano (HKY) pairwise sequence divergence was calculated in GENEIOUS PRO 5.4 software (Drummond *et al.*, 2009) for the mitochondrial *cytb* gene, which is widely used in phylogeographical studies.

Dispersal dynamics and ancestral area reconstruction

Ancestral areas were reconstructed to examine the biogeographical history of sea snakes. Three oceanic regions/ancestral areas were recognized based on other studies (VLIZ, 2009) which considered dispersal barriers (e.g. deep-sea trenches) and patterns of endemism and species ranges replicated across separate taxa. The three regions (Fig. 1 inset map) are the (1) Indian Ocean, (2) SE Asia (comprising *c.* 70% of the IAA) and (3) Australasia (which includes the eastern end of the IAA). Ancestral area reconstructions (AARs) were performed using the dated consensus tree (from the MRBAYES analysis) using Bayesian inference in BEAST 1.8 (Drummond & Rambaut, 2007), parsimony as implemented in MESQUITE 2.75 (Maddison & Maddison, 2009), and maximum likelihood as implemented in LAGRANGE (Ree & Smith, 2008). For all analyses, each sample (tip) was assigned to one of the three oceanic regions based on the collection locality (See Appendix S1 in Supporting Information for details of all analyses).

The BEAST analyses implemented novel methods to test whether rates of dispersal varied across lineages (clades) and/or events: the most appropriate model, selected using Bayes Factors, was adopted for AAR (see above) (See Appendix S3 for BEAST XML file). To test the importance of lineage-specific dispersal rates, we compared a model where different lineages (clades) were permitted different rates (using a 'random local clock' or RLC) (Drummond & Suchard, 2010) to a simpler model, which assumed a uniform dispersal rate across all lineages (a 'strict clock'). To test whether certain dispersal events were more likely, we tested four dispersal models of decreasing complexity: (1) a 'time-irreversible' model which assumed that all six dispersal events occurred at six different rates (Australasia → SE Asia; Indian Ocean → SE Asia; Australasia → Indian Ocean and the reverse), (2) a 'time-reversible' model which assumed three such rates (Australasia ↔ SE Asia; Indian Ocean ↔ SE Asia; Australasia ↔ Indian Ocean) and (3) a single-rate 'unordered' model which assumed a single common rate for all six events. We further evaluated (4) a single-rate 'ordered' model, which permitted only dispersals between adjacent regions (Australasia ↔ SE Asia; Indian Ocean ↔ SE Asia). There is no direct continental shelf connection between Australasia and the Northern/Western Indian Ocean, hence the 'ordered' model evaluates the hypothesis that sea snakes (with the possible exception of the pelagic, planktonic *H. (Pelamis) platurus*) moving between these regions must generally pass through SE Asia. In all models, a posterior probability of > 0.7 for a region for a node was considered
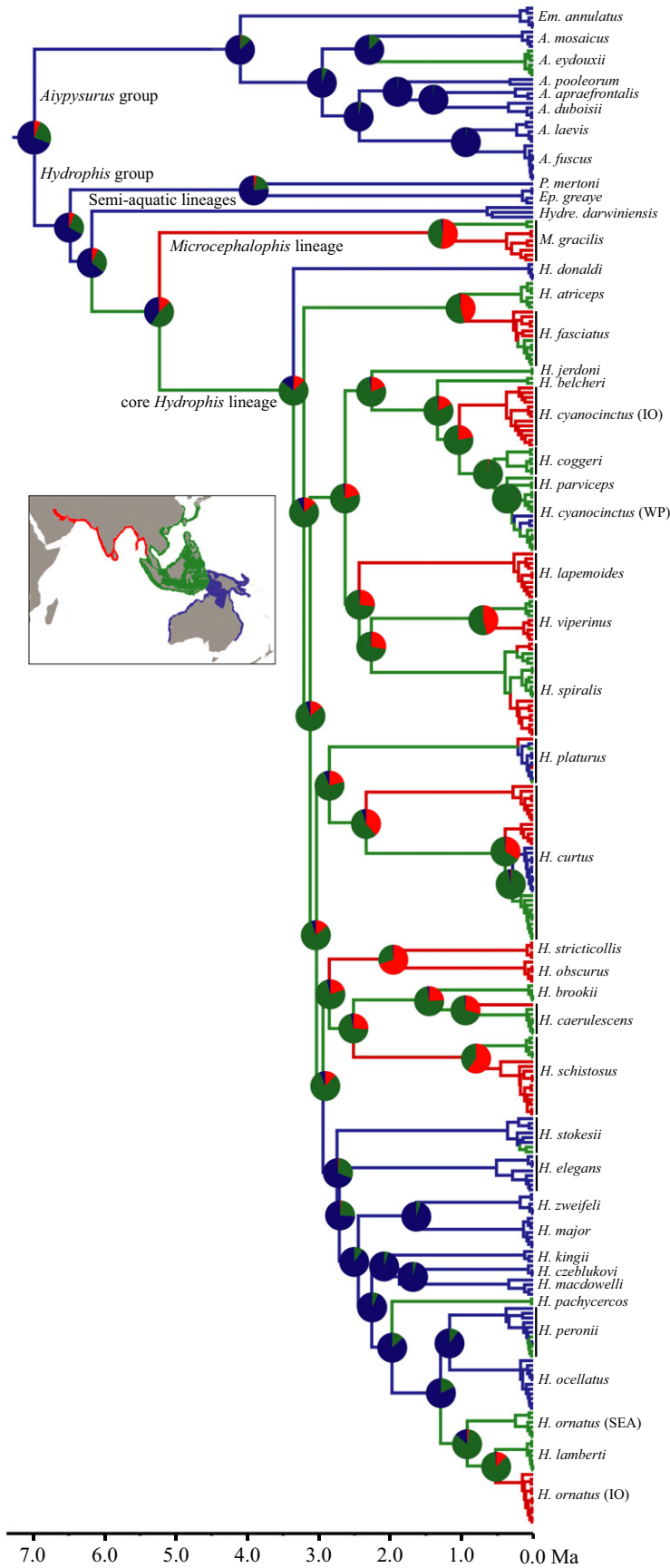
Figure 1 Time-calibrated tree of viviparous sea snakes, with Bayesian (BEAST) ancestral area reconstructions. Time-scale is in million years before present (Ma). Colours of the branches indicate the ancestral area reconstructions and correspond to the biogeographical/ancestral regions shown in map (Red: Indian Ocean (IO), Green: SE Asia (SEA), Blue: Australasia). Pie charts depict the relative posterior probability of the alternative ancestral areas for each node (WP – West Pacific, includes both SE Asia and Australia). See Fig. S2 in Supporting information for clade support values.

as strong support. These analyses used Markov chain Monte Carlo (MCMC) method to sample reconstructions in proportion to their probability and recorded the exact number of each of the six dispersal events in each sampled reconstruction (inferring event numbers using consensus node reconstructions will underestimate events if there are often multiple events along single long branches). In addition to using Beast to comparing these four event-specific models under a RLC, we also tested the fit of these four models in Bayestraits (Pagel et al., 2004), assuming a uniform dispersal rate across lineages (Bayestraits does not implement a RLC to accommodate lineage-specific dispersal rates).

The parsimony analyses used Mesquite 2.75 (Maddison & Maddison, 2009), and optimized regions and dispersals on the tree using an 'ordered model' (model number 4), which was the best-supported model identified in model testing (see above).

Maximum likelihood was implemented in the dispersal–extinction–cladogenesis (DEC) model in Lagrange (Ree & Smith, 2008) with ordered and unordered dispersal models. Likelihood ratio tests on Lagrange did not strongly favour either model, but both models produced generally similar results. Thus, we provide only the results of the ordered model, which is favoured in the Bayesian analyses and is biologically most reasonable (see above). At each node, range inheritance scenarios > 2 log-likelihood units better than all other possible scenarios were considered as strong support.

Although the Hydrophis sea snakes are among the most rapidly speciating tetrapods known (Sanders et al., 2010), this diversity was insufficient to permit statistical tests of relationship between geographical areas and speciation rate (BiSSE, GeoSSE), with robust results requiring 'roughly one or two hundred tip species' (Goldberg et al., 2011).

RESULTS

Phylogeny and divergence time estimates

ML (undated) and Bayesian (dated) analyses of the concatenated alignment recovered similar topologies, relative branch lengths and levels of support (Fig. S2 in Appendix S1 in Supporting Information). Both our ML and Bayesian analyses strongly recovered every sampled species except the Hydrophis ornatus complex as monophyletic (posterior probabilities (PP) > 0.9 and bootstrap values (BS) > 70%) (Fig. S2 in Appendix S1 in Supporting Information). Both analyses strongly recovered (PP > 0.9 and BS > 70%) reciprocally monophyletic clades within species that correspond to Indian Ocean versus SE Asian/West Pacific populations for Microcephalophis (Hydrophis) gracilis, Hydrophis caerulescens, H. (Lapemis) curtus, H. (Enhydrina) schistosus and H. (Thalassophina) viperinus (Fig. S2 in Appendix S1 in Supporting Information). Hydrophis curtus showed further population divergence with distinct clades in Phuket, Thailand (Indian Ocean), SE Asia and Australasia. The analysis also recovered distantly related cryptic lineages of H. cyanocinctus

and H. ornatus with allopatric distributions in the Indian Ocean or West Pacific/SE Asia (Fig. S1 in Appendix S1 in Supporting Information). However, the widely distributed species H. (Astrotia) stokesii, H. (Acalyptophis) peronii and H. (Pelamis) platurus did not display clear geographical genetic structure.

Divergence time estimates indicate that the speciation of the Aipysurus clade (containing the species of the genera Aipysurus and Emydocephalus) and the core Hydrophis clade (containing the species of the genus Hydrophis sensu Sanders et al., 2013a) each commenced c. 3.5 Ma (Aipysurus: 5.002–2.922 95% HPD; Hydrophis: 4.130–2.285 95% HPD) (Fig. 1, Fig. S2 in Appendix S1 in Supporting Information). However, the majority of the divergence time estimates between sister species and sister lineages (within species) ranged from 2.34 to 0.53 Ma (2.878–0.343 95% HPD) indicating a rapid late Pliocene or Pleistocene diversification (Table 1).

Corrected pairwise genetic (cytb) distances between sister lineages in the Indian Ocean and SE Asia ranged between 9.96–2.36%, and for sister lineages in Australasia and SE Asia ranged between 0.72–0.78% (Table 1). This was again consistent with a late Pliocene–Pleistocene speciation with respect to the estimated pairwise substitution rate of 3.3% per million years for the cytb gene in Hydrophiinae (Sanders et al., 2013a).

Dispersal dynamics and ancestral area reconstruction

The best-fitting model, as evaluated in Beast, allowed lineage-specific dispersal rates, and permitted dispersal only between adjacent regions ('ordered' model), with a single

Table 1 Percentage pairwise corrected genetic divergences, and mean divergence times (millions of years) between sister species/lineages in different Ocean basins.

Species/Lineage	Genetic divergence (corrected; %)	Mean divergence time (Ma)	Divergence Time (95% HPD, Ma)
A. eydouxii-A. mosaicus	7.10–7.39	2.297	2.878–1.679
H. atriceps-H. fasciatus	2.02–2.92	1.027	1.366–0.654
H. caerulescens (IO-SEA)	2.36–2.91	0.965	1.292–0.651
H. curtus (IO-WP)	8.64–9.96	2.337	2.895–1.698
H. curtus (SEA-AUS)	0.72–0.78	0.289	0.411–0.174
H. cyanocinctus (IO-WP)*	4.01–4.96	–	–
H. ornatus (IO-SEA)*	3.33–4.12	–	–
H. schistosus (IO-SEA)	4.05–4.96	0.716	0.967–0.490
H. lamberti-H. ornatus (IO)	1.04–1.30	0.526	0.718–0.343
H. viperina (IO-SEA)	4.05–4.85	0.708	0.977–0.457
M. gracilis (IO-SEA)	4.53–5.44	1.270	1.756–0.841

*These species are each currently considered single species. However, molecular analyses indicate that each consist of two cryptic lineages that do not show a sister species/lineage relationship (hence divergence time is not shown).
IO – Indian Ocean, SEA – SE Asia, AUS – Australasia, WP – West Pacific (includes both SEA and AUS).

Table 2 Inferred dispersal events from the three ancestral area reconstruction methods (A–C) and the fit of alternative dispersal models (D), which assume uniform or variable dispersal rates across lineages (clades) and across events. In the BEAST table (A), the actual numbers of events in the individual MCMC samples are listed first; the events 'inferred' by only examining nodal reconstructions in the Bayesian consensus tree are shown in parentheses.

From\To	Australasia	SE Asia	Indian Ocean
A: BEAST (variable rates across lineages, ordered)			
Australasia	–	17.1 (9)	*
SE Asia	18.2 (5)	–	17.7 (4)
Indian Ocean	*	11.6 (13)	–
B: Parsimony (ordered)			
Australasia	–	5	*
SE Asia	4	–	7
Indian Ocean	*	1	–
C: Lagrange (ordered, interspecific events only)			
Australasia	–	4	*
SE Asia	3	–	2
Indian Ocean	*	0	–

Dispersal models	−LognL	Bayes Factor
D: Fit of alternative dispersal models in BEAST		
Variable rates across lineages, 1 event rate (ordered)†	113.506	0 (best)
Variable rates across lineages, 1 event rate (unordered)	121.073	−15.134
Variable rates across lineages, 3 event rates (reversible)	117.398	−7.784
Variable rates across lineages, 6 event rates (irreversible)	118.378	−9.744
Uniform rates across lineages, 1 event rate (ordered)	122.758	−18.504

*Fixed to zero (see model testing in Appendix S1).

†The preferred model (number 4 in main text) assumes variable dispersal rates across lineages (RLC), and a common rate for all dispersal types, and also that dispersals are only possible between adjacent regions (i.e. ordered). See Appendix S1 for full description.

common rate for all four possible dispersal events (Australasia ↔ SE Asia; Indian Ocean ↔ SE Asia) (Table 2). Dispersal rates are relatively similar across most lineages, but planktonic *H. platurus* exhibits great (*c.* eightfold = 2.38) increase in dispersal rate compared to other sea snakes (Figs. 1, 2, S3 in Appendix S1 in Supporting Information; see below). BAYESTRAITS, which tested the four alternative event-specific dispersal models but under the assumption of a common dispersal rate across lineages, could not distinguish between the 'ordered', 3-rate and 6-rate models (all ΔBF < 5 compared to best model) but rejected the unordered model (ΔBF = 14.1).

All three AAR methods (Bayesian, parsimony, DEC: Figs. 1, 2, S3 in Appendix S1 in Supporting Information) recover an Australasian origin [Most recent common ancestor (MRCA)] for viviparous sea snakes, *c.* 6.9 Ma. Similarly, all three analyses indicate that the *Aipysurus* group also originated in Australasia, and subsequently diverged mostly

within this region. Parsimony, Bayesian and DEC analyses support an Australasian origin for the two semi-aquatic lineages. BEAST analyses indicated (probability = 0.73) a SE Asian origin for the MRCA of the core *Hydrophis* group, which accounts for *c.* 75% of extant species richness. Parsimony and DEC analyses are consistent with either an Australasian or SE Asian origin for this group. DEC analysis estimated an overall dispersal rate of 0.156 events per lineage per Myr (and an extinction probability of 0.016 per Myr) whereas BEAST analyses suggest dispersal rates ranging from 0.31–0.34 per lineage per my in most lineages, up to 2.38 in *H. platurus*.

DISCUSSION

Our time-calibrated molecular phylogenetic analyses and AARs reveal that although viviparous sea snakes had their origins in Australasia, they underwent rapid speciation after colonizing SE Asia during the last 3 million years. Phylogenetic analyses further recover reciprocally monophyletic clades that correspond to Indian Ocean versus SE Asian/West Pacific populations of five species of sea snakes indicating cryptic lineage diversity. AARs suggest that most of the SE Asian or the Indo-Australian Archipelago sea snake diversity is the result of *in situ* speciation. We discuss these findings here with reference to the geo-climatic history of the region, dispersal dynamics and the origins of IAA marine biodiversity.

Divergence times, sea snake speciation and sea level changes

Our findings are consistent with previous studies that showed an accelerated rate of speciation in the core *Hydrophis* radiation, with other viviparous sea snakes and their terrestrial sister groups having a slower background rate (Sanders *et al.*, 2010). The recency of many speciation events is consistent with Pleistocene vicariance. The dated tree (Fig. S2 in Appendix S1 in Supporting Information) suggests that the majority of speciation events in both the *Aipysurus* lineage and the core *Hydrophis* group have occurred since *c.* 3 Ma; this is also broadly consistent with corrected pairwise genetic (*cytb*) distances between sister lineages, which are typically < 6%, even for sister lineages spanning different oceans (see Table 1). Cyclic sea level changes that generated and dissolved barriers to dispersal between marine basins during the last 2.5 million years in the IAA (Voris, 2000; Lambeck *et al.*, 2002) are believed to have facilitated speciation of marine fauna via vicariance in isolated marine basins (e.g. De Bruyn & Mather, 2007; Crandall *et al.*, 2008). This may have been especially effective in sea snakes given that they are viviparous and thus lack the highly dispersive, planktonic larval stage that is expected to facilitate gene flow and population connectivity in many marine taxa.

The temporal diversification patterns uncovered for Indo-Pacific sea snakes are also consistent with species of marine invertebrates (Lavery *et al.*, 1996; Williams & Benzie, 1998;
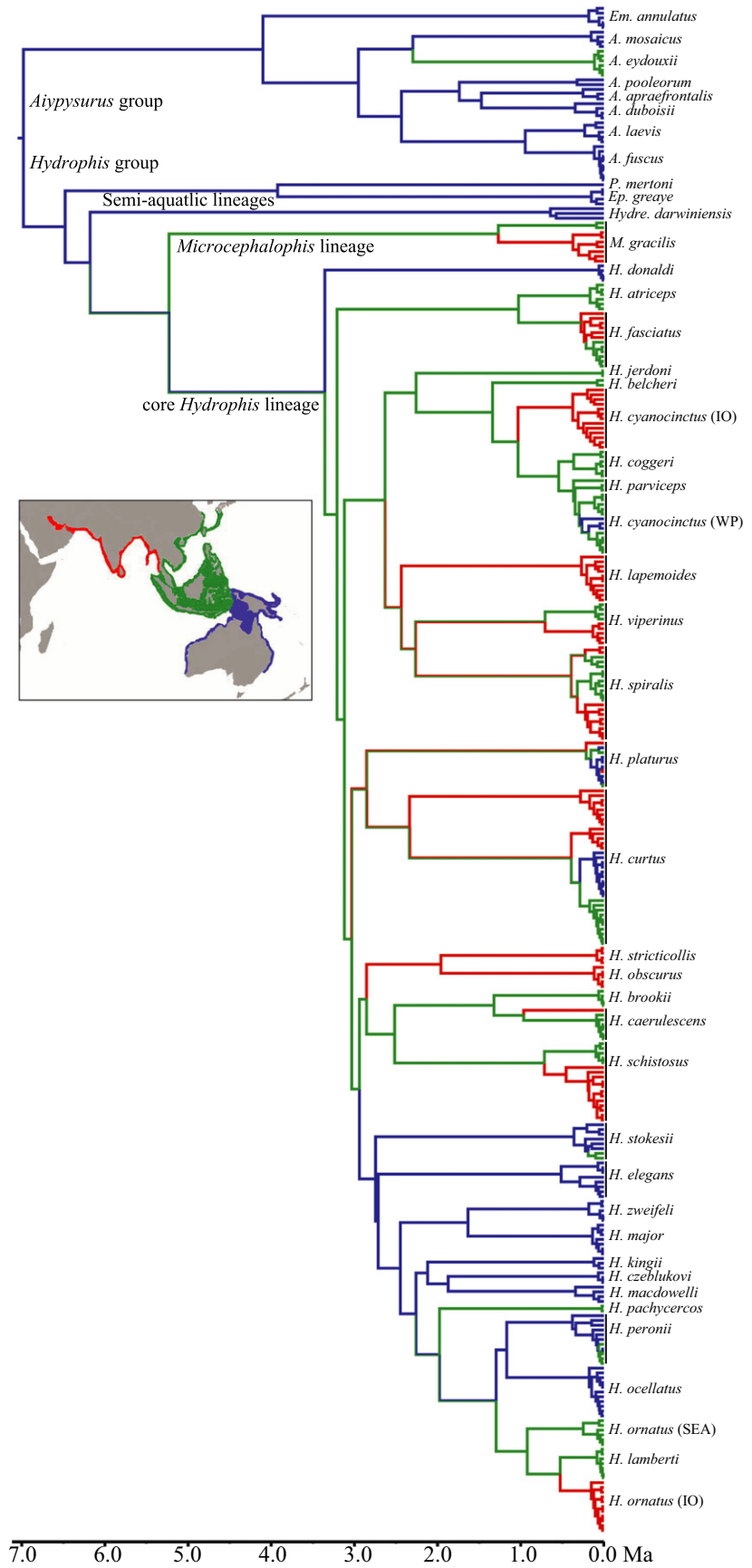
Figure 2 Time-calibrated tree of viviparous sea snakes, with parsimony ancestral area reconstructions. Time-scale is in millions of years before present (Ma). Colours of the branches indicate the most parsimonious ancestral area reconstructions for the node at the younger end, and correspond to the biogeographical/ancestral regions shown in map (Red: Indian Ocean, Green: SE Asia, Blue: Australasia). Two colours (e.g. in the core *Hydrophis* branch) indicate 2 equally-parsimonious reconstructions. See Fig. S2 in Supporting information for clade support values.

Benzie, 1999; Duda & Palumbi, 1999) and fish (Timm *et al.*, 2008; Drew & Barber, 2009; Leray *et al.*, 2010; Gaither *et al.*, 2011; Tornabene *et al.*, 2015) studied in this region. However, studies on other marine taxa from this region indicate that many taxa currently recognized as species pre-date the Pleistocene and potentially represent complexes of cryptic species (Barber & Bellwood, 2005; Renema *et al.*, 2008; Williams & Duda, 2008; Cowman & Bellwood, 2013). Thus, while Pleistocene vicariance has demonstrably played an important role in generating species and genetic diversity in many Indo-Pacific marine taxa, its contribution to total alpha diversity remains uncertain due to inadequate knowledge of species boundaries and thus, total species numbers. The current work and previous work has identified candidate new (cryptic) species in sea snakes (e.g. Ukuwela *et al.*, 2014), but this unappreciated alpha diversity is likely to be most prevalent in less studied groups such as many invertebrates. Comprehensive taxonomic revisions that incorporate dense molecular sampling from populations up to higher taxon clades are thus needed to clarify the diversification history and conservation status of marine groups in the IAA biodiversity hotspot.

Historical biogeography of Indo-Pacific sea snakes

Ancestral Area Reconstruction methods recover an Australasian origin for viviparous sea snakes, *c.* 6.9 million years ago. Similarly, AARs indicate that the *Aipysurus* group also originated in Australasia, and speciated mainly within this region. Of the *Aipysurus* group species, only the specialist fish egg-eaters *Emydocephalus ijimae, E. szczerbaki* (not sampled here) and *A. eydouxii* have colonized SE Asia and none have expanded into the Indian Ocean beyond the coast of Western Australia. Beast analyses recovered a SE Asian origin for the core *Hydrophis* group, and all three AAR methods indicated that subsequent diversification in this rapidly speciating clade occurred primarily in SE Asia, with subsequent dispersals into the Indian Ocean and re-colonization of Australasia. In the Beast AAR (Fig. 1), for instance, there are 34 divergences between lineages older than 0.5 Ma (candidate speciation events); 22 of these have > 0.7 posterior probabilities of occurring in SE Asia, 10 in Australasia, and 2 in the Indian Ocean (Figs 1–3). This suggests that most of the sea snake diversity in the SE Asia is derived from a period of rapid *in situ* diversification. Thus, although viviparous sea snakes originated in Australasia, SE Asia (which comprises most of the IAA) appears to be their primary 'centre of speciation'.

The best-fitting model evaluated in Beast AAR favoured lineage-specific dispersal rates, and permitted dispersal only between adjacent regions (Table 2). This best-fitting model implies no significant bias in direction of dispersal: thus, contrary to predictions of the overlap or accumulation models, taxa are not more likely to disperse into, rather than out of, SE Asia and thus the IAA. Viviparous sea snakes therefore provide little support for the 'region of accumulation hypothesis': there are few instances of peripheral speciation
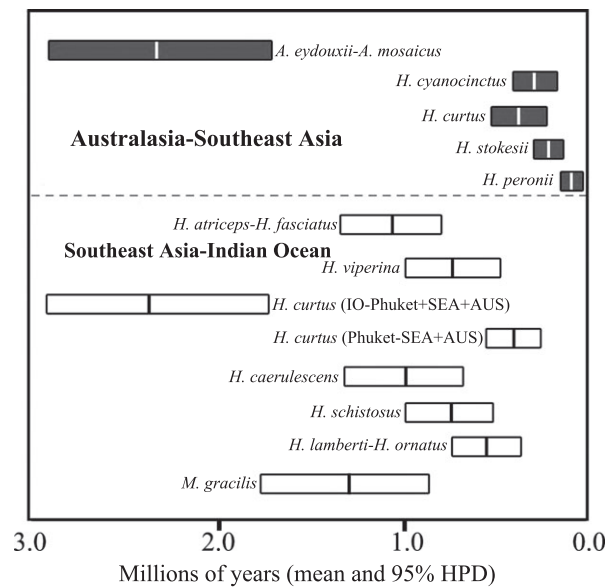
Figure 3 Divergence times (mean and 95% HPD intervals) between pairs of sister allopatric lineages, spanning Australasia (AUS) and SE Asia (SEA) (5 pairs, grey bars), and spanning SE Asia and the Indian Ocean (IO) (8 pairs, white bars). Most divergence events occurred in the last million years in both cases.

followed by subsequent re-colonization of SE Asia. Peripheral speciation is here identified as cladogenesis where one of the two resultant lineages is inferred to have (primitively) a SE Asian distribution and the other lineage to have (primitively) an external (Australasian or Indian Ocean) distribution. Across the entire tree, nodal reconstructions from Beast, Parsimony, and DEC analyses indicated two such speciation events between Australasia and SE Asia (*A. mosaicus-A. eydouxii* and within *H. curtus*) and six such events between the Indian Ocean and SE Asia (*H. ornatus-H. lamberti* and within *M. gracilis, H. caerulescens, H. curtus, H. schistosus,* and *H. viperinus*) (Figs 1–3 & S3 in Appendix S1 in Supporting information). These findings support a role of geographical/historical isolation at the periphery of the IAA in generating overall species/genetic diversity (Ladd, 1960). However, these events do not increase diversity in SE Asia (i.e. the IAA): the ancestral lineage of each species pair is inferred to be from SE Asia, the peripheral speciation event thus adds a new species to the diversity in the adjacent area (Australasia or Indian Ocean), but there is no evidence of secondary range expansion of these extralimital species back into SE Asia.

A small proportion of the sea snake diversity in SE Asia/IAA is consistent with the 'overlap' model: speciation entirely outside of SE Asia and subsequent re-colonization. When nodal reconstructions are examined in all three AAR methods, the only major external contribution appears to be from the *H. ornatus* clade (*H. stokesii, H. pachycercos, H. peroni, H. ornatus, H. ocellatus, H. lamberti*: sensu Sanders *et al.*, 2013a); a few lineages from this predominantly Australasian clade have secondarily extended their ranges back into SE

Asia (*H. stokesii*, *H. pachycercos*, *H. peroni* and the *H. ornatus*-*H. lamberti* clade). The Indian Ocean fauna has made little or no contribution to the SE Asian sea snake diversity (the only possible re-colonizations involve *H. fasciatus* and *H. spiralis*). The majority of sampled Indian Ocean species and lineages have a SE Asian origin and the regional sea snake fauna seems to be mainly derived from direct dispersal from SE Asia, with few dispersals in the other direction. These findings indicate that considerable speciation occurs outside of the IAA; however, subsequent inward dispersal into the IAA is not a major driver of species richness there.

Consistent with the inferences from nodal reconstructions above, all analyses suggested overall dispersals between SE Asia and Australasia occurred at the same frequencies in both directions (Table 2). The BEAST analyses suggested that dispersals between SE Asia and the Indian Ocean also occurred at approximately the same frequency in both directions; however, parsimony and DEC analyses indicated that dispersals from SE Asia to the Indian Ocean were more frequent than the reverse. However, the DEC analysis reconstructed very few events in total, by only considering events between rather than within species. The comparatively slower overall dispersal rate inferred in the DEC analysis might be due to the fact that it only evaluates rates in interspecific branches (the numerous recent dispersals on intraspecific branches were not considered). Alternatively, the broad (flat) prior in the Beast analysis might have allowed fast rates (see Appendix S1 in Supporting information). Dispersal rates were very similar across most lineages (0.31–0.34) with the exception of *H. platurus* (2.38). The relatively high dispersal rates seen in *H. platurus* likely reflect this species' unique ecology: *H. platurus* is the only species of sea snake with pelagic, planktonic habits (drifting with surface and subsurface currents) and consequently has the largest distribution of any squamate reptile (Heatwole, 1999).

According to the centre of refuge model, the proximity to stable habitats during Quaternary glacio-eustatic sea level changes (Voris, 2000; Woodruffe, 2003) was a major determinant of species survival, enabling re-colonization of unstable shallow water habitats through exportation from the source (Pellissier *et al.*, 2014). Distance to stable habitats (source populations) might be especially important for the maintenance of sea snake diversity in peripheral marine habitats due to their limited dispersal capabilities and reliance on shallow water habitats. Indeed, the most severe known local extinctions of sea snakes have occurred in the very remote Timor Sea reefs (Lukoschek *et al.*, 2013). This scenario is harder to evaluate with molecular trees, as the prime driver (elevated extinction outside biodiversity hotspots) is difficult to estimate using living species alone (Rabosky, 2010). However, some of our patterns discussed above as being consistent with the centre of origin model would also fit the centre of refuge model.

An evolutionary history where taxa which leave the IAA are rapidly 'pruned' by extinction would generate a phylogeny where most (inferred) speciation events are in the IAA and all the oldest clades are in the IAA, with subsequent and recent colonization of the Indian Ocean and Australasia. As discussed above, most (inferred) speciation events are in the IAA and the core *Hydrophis* lineage has its ancestral distribution in the IAA, and with multiple subsequent colonizations of the Indian Ocean and Australasian regions (Fig. 1). The broadly similar phylogenetic patterns expected by the 'centre of origin' and 'centre of refuge' models make them difficult to distinguish. However, the latter model would predict similar speciation rates and high diversity for all old clades (regardless of refuge region). The sea snake phylogeny here suggests long-term persistence (by itself) is not sufficient to generate high diversity, as the three most basal clades of sea snakes each have even longer inferred histories than the core *Hydrophis* group (though in the Australasian region: Fig. 1), yet have each attained only low to moderate diversity. However, huge phylogenies (several hundred taxa: Goldberg *et al.*, 2011; Davis *et al.*, 2013) are required to properly tease apart the effects of elevated *in situ* speciation versus higher extralimital extinction; this is in excess of the available species diversity of many relevant clades (including sea snakes, < 70 described species).

CAVEATS

Incomplete taxon sampling can affect biogeographical reconstructions and inferred dispersal patterns (Turner *et al.*, 2009). In this study, *c.* 70% of viviparous sea snake species were sampled: sampling was more complete for Australasian and Indian Ocean taxa (both > 75%), but less complete for SE Asia (< 60%). This would tend to bias results against reconstructing SE Asian range for ancestral nodes. Despite this potential bias, our AARs nevertheless recovered a SE Asian distribution for all basal, and most subsequent, speciation events in the core *Hydrophis* group. Hence, the importance of SE Asia as a centre of speciation for viviparous sea snakes is likely to remain and perhaps be amplified with additional species sampling. Similarly, two species (*H. coggeri* and *H. caerulescens*) were not sampled in one of the geographical areas they are known to occur (Australia). However, both species are highly nested in the *Hydrophis* clade so that their intraspecific relationships and distributions are unlikely to significantly impact the AARs at deeper nodes, including the initial diversification of the rapid *Hydrophis* radiation.

CONCLUSIONS

The drivers of the elevated diversification rate in the core *Hydrophis* group still need to be identified. They could involve extrinsic (geographical) factors, such as the formation of transient barriers (Palumbi, 1994) and proximity to habitat refugia (Pellissier *et al.*, 2014) in the Plio-Pleistocene, or intense competition (Briggs, 2005; Bowen *et al.*, 2013), or divergent selection in a highly heterogeneous and biodiverse environment (Rocha & Bowen, 2008). They could also be intrinsic: a recent study has suggested that plasticity of head size evolution contributed to rapid speciation in one clade

within this group (Sanders *et al.*, 2013b). Evaluation of whether the core *Hydrophis* group exhibits different diversification rates in different regions would answer this question, but robust inferences would require far more species than exist: at least 100–200 (Goldberg *et al.*, 2011) or > 300 (Davis *et al.*, 2013). However, pooling phylogenies of sea snakes and other vertebrate groups (fish) spanning this region might provide sufficient sample size (Goldberg *et al.*, 2011), although even with sufficiently large taxon sets, current implementations of these methods are highly problematic (Rabosky & Goldberg, 2015).

Distinguishing alternative diversification scenarios for the origins and maintenance of extraordinary marine biodiversity in the IAA remains a central goal in marine biogeography. Analyses of viviparous sea snakes suggest that SE Asia, which includes most of the IAA, has functioned mainly as a 'centre' or 'cradle' of speciation for viviparous sea snakes: the core *Hydrophis* group underwent rapid and largely *in situ* diversification during the last 3 Ma in SE Asia. Speciation either at the periphery (or outside) of SE Asia, followed by biased inwards range shifts, does not appear to be an important contributor of marine snake biodiversity of SE Asia and the IAA.

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REFERENCES

Allen, G.R. (2008) Conservation hotspots of biodiversity and endemism for Indo-Pacific coral reef fishes. *Aquatic Conservation: Marine and Freshwater Ecosystems*, **18**, 541–556.

Barber, P.H. & Bellwood, D.R. (2005) Biodiversity hotspots: evolutionary origins of biodiversity in wrasses (*Halichoeres*: Labridae) in the Indo-Pacific and new world tropics. *Molecular Phylogenetics and Evolution*, **35**, 235–253.

Barber, P.H., Palumbi, S.R., Erdmann, M.V. & Moosa, M.K. (2000) A marine Wallace's line? *Nature*, **406**, 692–693.

Barber, P.H., Erdmann, M.V., Palumbi, S.R. & Ayre, D. (2006) Comparative phylogeography of three codistributed stomatopods: origins and timing of regional lineage diversification in the Coral triangle. *Evolution*, **60**, 1825–1839.

Bellwood, D.R. & Hughes, T.P. (2001) Regional-scale assembly rules and biodiversity of coral reefs. *Science*, **292**, 1532–1535.

Benzie, J.A.H. (1999) Major genetic differences between crown-of-thorns starfish (*Acanthaster planci*) populations in the Indian and Pacific Oceans. *Evolution*, **53**, 1782–1795.

Bernardi, G., Bucciarelli, G., Costagliola, D., Robertson, D.R. & Heiser, J.B. (2004) Evolution of coral reef fish *Thalassoma* spp. (Labridae). 1. Molecular phylogeny and biogeography. *Marine Biology*, **144**, 369–375.

Bowen, B.W., Rocha, L.A., Toonen, R.J. & Karl, S.A. (2013) The origins of tropical marine biodiversity. *Trends in Ecology and Evolution*, **28**, 359–366.

Briggs, J.C. (1999) Coincident biogeographic patterns: Indo-West Pacific Ocean. *Evolution*, **53**, 326–335.

Briggs, J.C. (2005) The marine East Indies: diversity and speciation. *Journal of Biogeography*, **32**, 1517–1522.

Carpenter, K. & Springer, V. (2005) The center of the center of marine shore fish biodiversity: the Philippine Islands. *Environmental Biology of Fishes*, **72**, 467–480.

Carpenter, K.E., Barber, P.H., Crandall, E.D., Ablan-Lagman, M.C.A., Ambariyanto, Mahardika, G.N., Manjaji-Matsumoto, B.M., Juinio-Menez, M.A., Santos, M.D., Starger, C.J. & Toha, A.H.A. (2011) Comparative phylogeography of the Coral Triangle and implications for marine management. *Journal of Marine Biology*, **2011**, Article ID 396982. doi:10.1155/2011/396982.

Cowman, P.F. & Bellwood, D.R. (2013) The historical biogeography of coral reef fishes: global patterns of origination and dispersal. *Journal of Biogeography*, **40**, 209–224.

Crandall, E.D., Frey, M.A., Grosberg, R.K. & Barber, P.H. (2008) Contrasting demographic history and phylogeographical patterns in two Indo-Pacific gastropods. *Molecular Ecology*, **17**, 611–626.

Davis, M., Midford, P. & Maddison, W. (2013) Exploring power and parameter estimation of the BiSSE method for analyzing species diversification. *BMC Evolutionary Biology*, **13**, 38.

De Bruyn, M. & Mather, P.B. (2007) Molecular signatures of Pleistocene sea-level changes that affected connectivity among freshwater shrimp in Indo-Australian waters. *Molecular Ecology*, **16**, 4295–4307.

Drew, J. & Barber, P.H. (2009) Sequential cladogenesis of the reef fish *Pomacentrus moluccensis* (Pomacentridae) supports the peripheral origin of marine biodiversity in the Indo-Australian archipelago. *Molecular Phylogenetics and Evolution*, **53**, 335–339.

Drummond, A.J. & Rambaut, A. (2007) BEAST: Bayesian evolutionary analysis by sampling trees. *BMC Evolutionary Biology*, **7**, 214.

Drummond, A. & Suchard, M. (2010) Bayesian random local clocks, or one rate to rule them all. *BMC Biology*, **8**, 114.

Drummond, A.J., Ashton, B., Cheung, M., Heled, J., Kearse, M., Moir, R., Stones-Havas, S., Thierer, T. & Wilson, A. (2009) *Geneious*. Biomatters Limited, Available at: http://www.geneious.com/).

Duda, T.F. & Palumbi, S.R. (1999) Population structure of the black tiger prawn, *Penaeus monodon*, among western

Indian Ocean and western Pacific populations. *Marine Biology*, 134, 705–710.

Eble, J.A., Toonen, R.J., Sorenson, L., Basch, L.V., Papastamatiou, Y.P. & Bowen, B.W. (2011) Escaping paradise: larval export from Hawaii in an Indo-Pacific reef fish, the Yellow Tang (*Zebrasoma flavescens*). *Marine Ecology Progress Series*, 428, 245–258.

Ekman, S. (1953) *Zoogeography of the sea*. Sidgwick & Jackson, London.

Elfes, C.T., Livingstone, S.R., Lane, A. *et al.* (2013) Fascinating and forgotten: the conservation status of marine elapid snakes. *Herpetological Conservation and Biology*, 8, 37–52.

Gaither, M.R. & Rocha, L.A. (2013) Origins of species richness in the Indo-Malay-Philippine biodiversity hotspot: evidence for the centre of overlap hypothesis. *Journal of Biogeography*, 40, 1638–1648.

Gaither, M.R., Toonen, R.J., Robertson, D.R., Planes, S. & Bowen, B.W. (2010) Genetic evaluation of marine biogeographical barriers: perspectives from two widespread Indo-Pacific snappers (*Lutjanus kasmira* and *Lutjanus fulvus*). *Journal of Biogeography*, 37, 133–147.

Gaither, M.R., Bowen, B.W., Bordenave, T.R., Rocha, L.A., Newman, S.J., Gomez, J.A., van Herwerden, L. & Craig, M.T. (2011) Phylogeography of the reef fish *Cephalopholis argus* (Epinephelidae) indicates Pleistocene isolation across the Indo-Pacific barrier with contemporary overlap in the Coral Triangle. *BMC Evolutionary Biology*, 11, 189.

Goldberg, E.E., Lancaster, L.T. & Ree, R.H. (2011) Phylogenetic inference of reciprocal effects between geographic range evolution and diversification. *Systematic Biology*, 60, 451–465.

Halas, D. & Winterbottom, R. (2009) A phylogenetic test of multiple proposals for the origins of the East Indies coral reef biota. *Journal of Biogeography*, 36, 1847–1860.

Heatwole, H. (1999) *Sea snakes*, 2nd edn. University of New South Wales Press, Sydney, Australia.

Hodge, J.R., Read, C.I., van Herwerden, L. & Bellwood, D.R. (2012) The role of peripheral endemism in species diversification: evidence from the coral reef fish genus *Anampses* (Family: Labridae). *Molecular Phylogenetics and Evolution*, 62, 653–663.

Hoeksema, B.W. (2007) Delineation of the Indo-Malayan centre of maximum marine biodiversity: the Coral Triangle. *Biogeography, time, and place: distributions, barriers, and islands* (ed. by W. Renema), pp. 117–178. Springer, Netherlands.

Hoskin, M.G. (1997) Effects of contrasting modes of larval development on the genetic structures of populations of three species of prosobranch gastropods. *Marine Biology*, 127, 647–656.

Hubert, N., Meyer, C.P., Bruggemann, H.J., Guérin, F., Komeno, R.J.L., Espiau, B., Causse, R., Williams, J.T. & Planes, S. (2012) Cryptic diversity in Indo-Pacific coral-reef fishes revealed by DNA-barcoding provides new support to the centre-of-overlap hypothesis. *PLoS ONE*, 7, e28987.

Hughes, T.P., Bellwood, D.R. & Connolly, S.R. (2002) Biodiversity hotspots, centres of endemicity, and the conservation of coral reefs. *Ecology Letters*, 5, 775–784.

Jackson, J.B.C., Jung, P., Coates, A.G. & Collins, L.S. (1993) Diversity and extinction of tropical American mollusks and emergence of the Isthmus of Panama. *Science*, 260, 1624–1626.

Kass, R.E. & Raftery, A.E. (1995) Bayes factors. *Journal of the American Statistical Association*, 90, 773–795.

Ladd, H.S. (1960) Origin of the Pacific island molluscan fauna. *American Journal of Science*, 258, 137–150.

Lambeck, K., Esat, T.M. & Potter, E.-K. (2002) Links between climate and sea levels for the past three million years. *Nature*, 419, 199–206.

Lavery, S., Moritz, C. & Fielder, D.R. (1996) Indo-Pacific population structure and evolutionary history of the coconut crab *Birgus latro*. *Molecular Ecology*, 5, 557–570.

Leray, M., Beldade, R., Holbrook, S.J., Schmitt, R.J., Planes, S. & Bernard, G. (2010) Allopatric divergence and speciation in coral reef fish: the three-spot dascyllus, *Dascyllus trimaculatus*, species complex. *Evolution*, 64, 1218–1230.

Lourie, S.A. & Vincent, A.C.J. (2004) A marine fish follows Wallace's Line: the phylogeography of the three-spot seahorse (*Hippocampus trimaculatus*, Syngnathidae, Teleostei) in Southeast Asia. *Journal of Biogeography*, 31, 1975–1985.

Lukoschek, V. & Keogh, J.S. (2006) Molecular phylogeny of sea snakes reveals a rapidly diverged adaptive radiation. *Biological Journal of the Linnean Society*, 89, 523–539.

Lukoschek, V., Waycott, M. & Marsh, H. (2007) Phylogeography of the olive sea snake, *Aipysurus laevis* (Hydrophiinae) indicates Pleistocene range expansion around northern Australia but low contemporary gene flow. *Molecular Ecology*, 16, 3406–3422.

Lukoschek, V., Scott Keogh, J. & Avise, J.C. (2012) Evaluating fossil calibrations for dating phylogenies in light of rates of molecular evolution: a comparison of three approaches. *Systematic Biology*, 61, 22–43.

Lukoschek, V., Beger, M., Ceccarelli, D., Richards, Z. & Pratchett, M. (2013) Enigmatic declines of Australia's sea snakes from a biodiversity hotspot. *Biological Conservation*, 166, 191–202.

Maddison, W.P. & Maddison, D.R. (2009) *Mesquite: a modular system for evolutionary analysis*. Available at: http://mesquiteproject.org).

Mironov, A.N. (2006) Centers of marine fauna redistribution. *Zoologicheskii Zhurnal*, 85, 3–17.

Mora, C., Chittaro, P.M., Sale, P.F., Kritzer, J.P. & Ludsin, S.A. (2003) Patterns and processes in reef fish diversity. *Nature*, 421, 933–936.

Pagel, M., Meade, A. & Barker, D. (2004) Bayesian estimation of ancestral character states on phylogenies. *Systematic Biology*, 53, 673–684.

Palumbi, S.R. (1994) Genetic divergence, reproductive isolation, and marine speciation. *Annual Review of Ecology and Systematics*, 25, 547–572.

Pellissier, L., Leprieur, F., Parravicini, V., Cowman, P.F., Kulbicki, M., Litsios, G., Olsen, S.M., Wisz, M.S., Bellwood, D.R. & Mouillot, D. (2014) Quaternary coral reef refugia preserved fish diversity. *Science*, **344**, 1016–1019.

Rabosky, D.L. (2010) Extinction rates should not be estimated from molecular phylogenies. *Evolution*, **64**, 1816–1824.

Rabosky, D.L. & Goldberg, E.E. (2015) Model inadequacy and mistaken inferences of trait-dependent speciation. *Systematic Biology*, **64**, 340–355.

Rambaut, A. & Drummond, A.J. (2007) *Tracer*. Available at: http://beast.bio.ed.ac.uk/Tracer/).

Randall, J.E. (1998) Zoogeography of shore fishes of the Indo-Pacific region. *ZOOLOGICAL STUDIES-TAIPEI-*, **37**, 227–268.

Rasmussen, A.R. (2001) Sea snakes. *Living marine resources of the western central Pacific* (ed. by K.E. Carpenter and V.H. Niem), pp. 3987–4008. Food and Agriculture Organization, Rome.

Rasmussen, A.R. (2002) Phylogenetic analysis of the "true" aquatic elapid snakes Hydrophiinae (sensu Smith et al., 1977) indicates two independent radiations to water. *Steenstrupia*, **27**, 47–63.

Ree, R.H. & Smith, S.A. (2008) Maximum likelihood inference of geographic range evolution by dispersal, local extinction, and cladogenesis. *Systematic Biology*, **57**, 4–14.

Renema, W., Bellwood, D.R., Braga, J.C., Bromfield, K., Hall, R., Johnson, K.G., Lunt, P., Meyer, C.P., McMonagle, L.B., Morley, R.J., O'Dea, A., Todd, J.A., Wesselingh, F.P., Wilson, M.E.J. & Pandolfi, J.M. (2008) Hopping hotspots: global shifts in marine biodiversity. *Science*, **321**, 654–657.

Rocha, L.A. & Bowen, B.W. (2008) Speciation in coral-reef fishes. *Journal of Fish Biology*, **72**, 1101–1121.

Ronquist, F. & Huelsenbeck, J.P. (2003) MRBAYES 3: Bayesian phylogenetic inference under mixed models. *Bioinformatics*, **19**, 1572–1574.

Sanders, K.L. & Lee, M.S.Y. (2008) Molecular evidence for a rapid late-Miocene radiation of Australasian venomous snakes (Elapidae, Colubroidea). *Molecular Phylogenetics and Evolution*, **46**, 1165–1173.

Sanders, K.L., Lee, M.S.Y., Leys, R., Foster, R. & Keogh, J.S. (2008) Molecular phylogeny and divergence dates for Australasian elapids and sea snakes (Hydrophiinae): evidence from seven genes for rapid evolutionary radiations. *Journal of Evolutionary Biology*, **21**, 682–695.

Sanders, K.L., Mumpuni & Lee, M.S.Y. (2010) Uncoupling ecological innovation and speciation in sea snakes (Elapidae, Hydrophiinae, Hydrophiini). *Journal of Evolutionary Biology*, **23**, 2685–2693.

Sanders, K.L., Lee, M.S.Y., Mumpuni, Bertozzi, T. & Rasmussen, A.R. (2013a) Multilocus phylogeny and recent rapid radiation of the viviparous sea snakes (Elapidae: Hydrophiinae). *Molecular Phylogenetics and Evolution*, **66**, 575–591.

Sanders, K.L., Rasmussen, A.R., Mumpuni, Elmberg, J., de Silva, A., Guinea, M.L. & Lee, M.S.Y. (2013b) Recent rapid speciation and ecomorph divergence in Indo-Australian sea snakes. *Molecular Ecology*, **22**, 2742–2759.

Santini, F. & Winterbottom, R. (2002) Historical biogeography of Indo-western Pacific coral reef biota: is the Indonesian region a centre of origin? *Journal of Biogeography*, **29**, 189–205.

Scanlon, J.D. & Lee, M.S.Y. (2011) The major clades of snakes: morphological evolution, molecular phylogeny, and divergence dates. *Reproductive biology and phylogeny of snakes* (ed. by R.D. Aldridge and D.M. Sever), pp. 55–95. CRC Press, Boca Raton, Florida.

Smith, M. (1926) *Monograph of the sea-snakes (Hydrophidae)*. Taylor and Francis, London, UK.

Stamatakis, A. (2006) RAxML-VI-HPC: maximum likelihood-based phylogenetic analyses with thousands of taxa and mixed models. *Bioinformatics*, **22**, 2688–2690.

Timm, J., Figiel, M. & Kochzius, M. (2008) Contrasting patterns in species boundaries and evolution of anemonefishes (Amphiprioninae, Pomacentridae) in the centre of marine biodiversity. *Molecular Phylogenetics and Evolution*, **49**, 268–276.

Tornabene, L., Valdez, S., Erdmann, M. & Pezold, F. (2015) Support for a 'Center of Origin' in the Coral Triangle: cryptic diversity, recent speciation, and local endemism in a diverse lineage of reef fishes (Gobiidae: Eviota). *Molecular Phylogenetics and Evolution*, **82**, Part A, 200–210.

Turner, A.H., Smith, N.D. & Callery, J.A. (2009) Gauging the effects of sampling failure in biogeographical analysis. *Journal of Biogeography*, **36**, 612–625.

Ukuwela, K.D.B., de Silva, A., Mumpuni, Fry, B.G. & Sanders, K.L. (2014) Multilocus phylogeography of the spine-bellied sea snake (Hydrophis curtus, Elapidae) reveals historical vicariance and cryptic lineage diversity. *Zoologica Scripta*, **43**, 472–484.

Veron, J.E.N. (1995) *Corals in space and time: the biogeography and evolution of the Scleractinia*. Cornell University Press, Ithaca.

VLIZ (2009) *Longhurst Biogeographical Provinces* Available at: http://www.marineregions.org/) (accessed 5 September 2013).

Voris, H.K. (2000) Maps of Pleistocene sea levels in Southeast Asia: shorelines, river systems and time durations. *Journal of Biogeography*, **27**, 1153–1167.

Williams, S.T. & Benzie, J.A.H. (1998) Evidence of a Biogeographic break between populations of a high dispersal Starfish: congruent regions within the Indo-West Pacific defined by color morphs, mtDNA, and allozyme data. *Evolution*, **52**, 87–99.

Williams, S.T. & Duda, T.F., Jr (2008) Did tectonic activity stimulate Oligo-Miocene speciation in the Indo-West Pacific? *Evolution*, **62**, 1618–1634.

Woodland, D.J. (1983) Zoogeography of the Siganidae (Pisces) – an interpretation of distribution and richness patterns. *Bulletin of Marine Science*, **33**, 713–717.

Woodruffe, D.S. (2003) Neogene marine transgressions, palaeogeography and biogeographic transitions on the Thai-Malay Peninsula. *Journal of Biogeography*, **30**, 551–567.

SUPPORTING INFORMATION

Additional Supporting Information may be found in the online version of this article:

Appendix S1 Supplementary Materials and Methods and supplementary Figures.

Appendix S2 Details of the specimens, voucher numbers and the respective Genbank accession numbers used in the molecular phylogenetic analysis.

Appendix S3 Aligned data set in Nexus format with MrBayes commands and Beast xml file for biogeographical reconstruction.

BIOSKETCH

Kanishka D.B. Ukuwela is a recent PhD graduate from the University of Adelaide, Australia, and now a Lecturer at Rajarata University of Sri Lanka. His current research is focused on the origins, evolution and systematics of the South Asian herpetofauna.

Author contributions: K.D.B.U., M.S.Y.L., K.L.S. conceived the study. K.D.B.U., A.R.R., AdS, Mu, B.G.F., P.G., M.R. and K.L.S. collected samples. K.D.B.U. and K.L.S. generated data. K.D.B.U., K.L.S. and M.S.Y.L. analysed data and K.D.B.U., M.S.Y.L. and K.L.S. wrote the paper.

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